February 25, 1958

Dr. Herman M. Kalckar
National Institutes of Health
Bethesda 14, Maryland

Dear Herman:

I was very glad to hear that you were able to straighten out the
question of travel expense. We are looking forward to seeing you here.
As a matter of fact the time during the meetings will be so busy that
there may be relatively little occasion for us to get together privately.
Would you consider staying over another day so that we might have some
chance for quiet discussion? There are a number of things I really want
to pin you down on.

Of course we are even more pleased to hear of the renewal of your
interests in the Gal problem. I realize of course that you are pre-
occupied by many other distractions and that having lost Kiyosha to
another program would not help the continuity of our collaboration.
However if you will look through the correspondence I think you will
see that we have had much too sketchy a picture of what has been going
on and too often we could get our first clear impressions only from the
rather condensed version of a hasty manuscript. Now that the most excit-
ing initial features have in a sense passed, I think that we can perhaps
settle down to a more considered discussion.

One point of confusion that has never been cleared up for me is the
extent of inducability of the various enzymes in wild type E. coli.
Kiyosha once promised to send me the quantitative data on this point but
never has. I am by no means very happy about the use of nutriant media
for the bulk of the experiments as I am sure this would obscure induc-
tion effects.

As I am sure you realize the mutant step has been worked on until
now represents the simplest cases and they of course did give the very
exciting result of correlating two genetic cistrons with two specific
enzymes. In spite of the speculation on such points elsewhere, I think
this was the first real verification of that idea. But Esther and I are
both a little concerned about too hasty a generalization, especially
when this is based on a selected sample of mutants. The Gal_3 case will
be an example. It is just as puzzling to us why this should seem to
belong to two cistrons as its enzymatic behavior is puzzling to you.
But until this is cleared up on both sides I think it stands as a serious
challenge to the simple hypothesis. Recently Esther sent you another

culture labeled Gal_9. This seems to be genetically analogous to Gal_3 but is distinct from it by recombination test. If it should prove to have the same biochemical behavior as Gal_3 it would indicate that this is a significant class of mutants and not an exception worthy of being ignored. It will then, it seems to me, be extremely important to find out what its defect is in the metabolism of galactose.

If we had not made it clear before the essential genetic information about Gal_3, (and the same for Gal_9) is this: it is a distinct mutant, different by recombination test from every other one tested. It is therefore unlikely to be a simple deletion such as Benzer has described, because such a type would be expected to be apparently the same, by recombination test, with a number of other mutants. Furthermore it does show occasional reverse mutation and therefore is equally unlikely to be a simple deletion. However when heterogenotes are made between Gal_3 and any of the mutants either from the Gal_1 or the Gal_2 cistrons, typical position effects are seen. This is, I think, contrary to any simple picture of the organization of the genetic material into discrete cistrons. It might help however to know what the enzymological effect is.

We have, as you know, at least 50 other mutants but we are not going to bother you with them unless we find some peculiarity that would warrant more detailed analysis. They are being systematically screened for their cistron behavior and we will let you know the picture if any further exceptions appear. We are also testing all of the mutants for the temperature sensitivity at least as far as their growth fermentative ability is concerned. We are also screening all of the existing mutants, and looking for a great many more, from the point of view of dependence on galactose, obviously in the hope that these might reflect the missing class of etimerase mutants.

Our failure to find etimerase mutants so far may be a consequence of the inability of such mutants to grow even on a medium containing galactose. Do you suppose there would be very much point in adding UDP Gal to our complete medium? If not this, can you suggest any other supplements which might be used instead? If we do use UDP Gal, do you suggest we use the commercial materials obtainable from the Sigma Company or can you suggest a better source?

Still another possibility, it seems to me, is that an etimerase mutant may not appear to be galactose negative on our usual test. Can you think of any by-pass mechanisms that would take the place of the etimerase? Of course there is the remaining possibility that UDP Gal might be quite toxic if accumulated inside the cell.

We will be very happy to have your comments on these points.

Do you remember W3142? We still do not have the last word on this but there is now some reason to believe that it is a double mutant. One of the factors may be a fairly typical Gal- linked in the other cistron. The other factor would either prevent efficient transduction to the strain or might be a modifier resulting in "slow" fermentation. The whole story is not at all finished but I thought you might want to know this current interpretation. In any case the publication last year was highly premature and I hope we can have more effective communication on both sides on such questions in the future.

I want to be sure I understood your last letter about the symposium expenses. Do you now have permission to receive payment from us, as we offered in the first place, or will you get payment direct from the NIH?

Can you tell me just what is the technical problem in the estimation of the epimerase of the Gal₃ mutant?

As ever, yours cordially,

Joshua Lederberg
Professor of Medical Genetics

JL/ew